REMARKS

Applicants thank Examiner Woitach for issuing the January 2, 2003 Office Action as a non-final action, thereby providing applicants a further opportunity to demarcate the presently claimed invention, by the claims as amended herein.

Applicants have requested the certified copy of the German application no. 196 31 357.0 filed on August 2, 1996, and will submit same to the Office upon receipt.

Rejection of Claims and Traversal Thereof

In the January 2, 2003 Office Action,

claims 14-25, 27, 29, 38-61, 65 and 66 were rejected under 35 U.S.C. §112, second paragraph;

claims 14-25, 27, 29, 38-61, 65 and 66 stand rejected under 35 U.S.C. §103 (a) as being unpatentable over Donnelly, et al. (WO 96/00583) and Johnson (U.S. Patent No. 5,658,785) in further view of Whittle, et al. (U.S. Patent No. 5,658,785);

claims 16, 18, 20 and 50 stand rejected under 35 U.S.C. §103 (a) as being unpatentable over Donnelly, et al. (WO 96/00583), Johnson (U.S. Patent No. 5,658,785) and Whittle, et al. (U.S. Patent No. 5,658,785) in further view of Gissmann, et al. (WO 96/11272); and

claim 61 stands rejected under 35 U.S.C. §103 (a) as being unpatentable over Donnelly, et al. (WO 96/00583), Johnson (U.S. Patent No. 5,658,785) and Whittle, et al. (U.S. Patent No. 5,658,785) in further view of Stanley, et al. (U.S. Patent No. 6,096,869).

These rejections are hereby traversed and reconsideration of the patentability of the pending claims is requested in light of the following remarks.

Rejection under 35 U.S.C. §112, second paragraph

Claims 14-25, 27, 29, 38-61, 65 and 66 were rejected under 35 U.S.C. §112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Applicants have amended the claims thereby obviating the rejections. Accordingly, applicants request that all rejections under 35 U.S.C. §112, second paragraph be withdrawn.

Rejection under 35 U.S.C. §103 (a)

In the January 2, 2003 Office Action, claims 14-25, 27, 29, 38-61, 65 and 66 were rejected under 35 U.S.C. §103 (a) as being unpatentable over Donnelly, et al. and Johnson, and in further view of Whittle, et al. Applicants submit that Donnelly, et al. in combination with Johnson and Whittle, et al. does not render applicants' claimed invention *prima facie* obvious.

The present invention relates to an adeno-associated virus vector comprising a nucleotide sequence encoding a fusion polypeptide. The fusion polypeptide comprises a structural papillomavirus polypeptide and a non-transforming E6 or E7 early papillomavirus polypeptide having the C-terminus of the structural papillomavirus polypeptide connected to the N-terminus of the non-transforming E6 or E7 early papillomavirus polypeptide. The term "non-transforming" is defined at page 4 in the amended specification, submitted on July 24, 2001, as "comprising any peptide or polypeptide, that is coded by an early papillomavirus gene (ORF) or fragment thereof, and is non-transforming by nature or through intervention." . . . Through intervention, the transforming ability of a (poly)peptide is destroyed by deleting a part of the ORF." It is further stated at page 4 that "[p]art of the E6-ORF has been deleted beforehand, so that the transforming properties of E6 were destroyed." Thus, applicants' vectors encode for a transformation[2]-deficient early papillomavirus polypeptide that reduces the risk of de novo tumor induction, yet can till trigger the immune system to mount an immune response against expressed antigens.

According to the Office the E6- or E7-ORFs were not considered transforming in the art.

[2] The term "transformation" is defined in applicants' specification, at the top of page 4, as "the conversion of a normal cell into a tumor cell which has the capacity for unlimited proliferation."

Specifically, the Office stated that:

> "As evidenced to this fact, Swan *et al.* (Arch Virol, 1994) teach that transfection 'of E6 and E7 genes from the oncogenic HPV types into primary rodent cells causes immortalization, but not transformation.' (page 110, second full paragraph)."

Thus, Swan states that the gene causes immortalization, but applicants are claiming a protein that is expressed by the E6 or E7 gene and rendered transformation-deficient by deleting part of the conserved sequence of the E6 or E7 gene that encodes for the transformation properties. The Swan article is ambiguous regarding whether the gene or expressed protein is being discussed. Clearly, the remainder of the sentence quoted by the Office discusses that transformation does occur, albeit, a little slower with some HPV types. Specifically, the full sentence states:

> "E6 or E7 genes from the oncogenic HPV types into primary rodent cells causes immortalization, but not transformation **until after a long latency, usually involving a crisis stage."** (emphasis added)

Further in the cited paragraph, there is some discussion that may explain the long latency by stating that "maybe the cells, . . . contain many cytogenetic abnormalities [22] and these may reflect the activation of oncogenes and other factors." Further explanation for the transformation process can be found at page 106, first full paragraph, of the Swan review article, wherein it specifically states:

> "The high-risk HPVs can transform established cell-lines [67] and in the presence of an activated oncogene HPVs bind to and inactivate several of the tumor-suppressors [16, 20, 64]. Integration of high-risk HPVs probably affects transformation by increasing E6/E7 levels, thus decreasing the activity of the tumor suppressors. The low risk HPVs have E6 and E7 proteins that bind tumor suppressors proteins with low affinity and transform much less efficiently."

Thus, the speed of the transformation process is determined by whether the HPV virus is a high or low risk virus. The section cited by the Office does not specifically state whether the E6 or E7 genes are from a high risk or low risk HPV. However, it is very clear that the encoded E6 and E7 proteins cause transformation by decreasing the activity of tumor suppressors, whether it occurs immediately or has a latency period.

Numerous articles reinforce the transforming properties of E6 and E7 proteins. For example, a primary source of molecular biology, The Encyclopedia of Molecular Biology, published in 1994, expressly states that E6 and E7 have transforming abilities. Specifically, at page 789, (a copy of which is included in Appendix A) the proteins are discussed as follows:

> "**E6,E7.** The major proteins encoded by papillomaviruses that are relevant to transformation are transcribed from the E6 and E7 ORFs."

Further, a 1995 article by Tommasino, et al. (copy in Appendix A) states that:

> "[E]xtensive studies on HPV E6 and E7 proteins have demonstrated their involvement in malignant transformation."

The abstract of Liu, et al. (copy in Appendix A) states that:

> "The transform potential of the human papillomavirus (HPV) type 16 has been defined largely in the E7, E6 and E5 oncoproteins, the major transforming capability residing in the E7 gene."

This abstract further states:

> "that the transforming potential is directly correlated to the expression levels of the oncoprotein and that a threshold level of the E7 oncoprotein may be required before the cells can be fully transformed. This supports the hypothesis that the transformation processes include at least two separate and continuous steps which first lead to immortalization and then to metastasis, in agreement with the clinical progression of genital tumors from benign to malignancy."

Thus, the E6 and E7 proteins encoded by the E6- and E7-ORF, respectively, are involved in transformation. The section cited by the Office in the Swan, et al. article discusses data from references that were published in 1988 and 1991. Clearly at that time, there was a lack of knowledge and understanding that the transformation process is correlated to the level of expression of the E6 and E7 proteins and that in fact the transformation process includes at least two separate and continuous steps, which first lead to immortalization and then to metastasis.

As stated above, Applicants' vectors comprise a transformation deficient early papillomavirus polypeptide encoded by E6 or E7 genes that have a section deleted therefrom that encodes for the transforming properties of the expressed protein. Advantageously, the use of the non-transforming

early papillomavirus polypeptide reduces the risk of de novo tumor induction, yet still triggers the immune system to mount an immune response against expressed antigens. Clearly, none of the cited references alone or in combination describe, teach or suggest applicants' claimed invention.

The Office has combined the teachings of Whittle, et al., Donnelly, et al., and Johnson in an attempt to establish a *prima facie* case of obviousness. Donnelly, et al. teaches DNA plasmids encoding for polypeptides of papillomavirus. However, if more that one polypeptide is used to induce an immune response, then **separate DNA plasmids** are used as discussed on page 8, in the third paragraph of the Donnelly, et al reference. Whittle, et al. teaches fused polypeptides of HPV and Johnson describes a eukaryotic expression system comprising recombinant Adeno-Associated Virus (AVV) vectors that are used in combination with a helper virus for transfection of cells.

Applicants submit that the proposed combination does not in any way disclose, teach or suggest each and every element of the presently claimed invention because none of the cited references either alone or in combination teach or suggest a **fused polypeptide** comprising a structural papillomavirus polypeptide encoded by an open reading frame selected from the group consisting of L1-ORF, L2-ORF and fragments thereof, and an early E6-ORF, E7-ORF or fragments thereof, **wherein a part of the nucleotide sequence of the E6-ORF or E7-ORF gene is deleted and the deleted part encodes for the transforming properties of an expressed protein.** By deleting a section of the E6-ORF or E7-ORF that encodes for the transforming properties, the expressed protein cannot bind to and inactivate tumor-suppressors, and thus, is rendered non-transforming.

None of the cited references provides any motivation to go in the direction of applicants' claimed invention. As stated above, Donnelly, et al. teaches the use of one gene per **non-viral** plasmid. Whittle, et al. does not in any way, disclose, teach or suggest deleting any part of the E6-ORF or E7-ORF gene before inclusion in the prokaryotic expression system. The full E6-ORF or E7-ORF gene is included and the reference is devoid of any discussion relating to a deletion of a part of the genes to render any expressed proteins as non-transforming. Johnson is only related to **viral** vectors and does not addresss E7 and E6 proteins or encoding genes.

It is incumbent on the Office to provide some suggestion or teaching in the prior art that would lead one skilled in the art to proceed in the direction of applicants' claimed invention. Applicants respectfully submit that the Office has not provided any objective or specific teachings or suggestions in the cited prior art to motivate one skilled in the art to modify any of the cited references. What is the asserted motivation put forth in any of the cited references to generate a viral vector comprising structural papillomavirus polypeptide and a transformation deficient E6 or E7 early papillomavirus polypeptide that was expressed from an **E6-ORF or E7-ORF gene having a section of the sequence deleted so that the expressed protein does not have transforming properties?** The Courts have addressed this issue numerous times and have stated that "[t]he mere fact that the prior art could be so modified would not have made the modification obvious unless the prior art suggested the desirability of the modification." *In re Mills*, 16 U.S.P.Q.2d 1430 (Fed. Cir. 1990). Thus, this allegedly "obvious" direction is supported only by the Office's reinterpretation of the art in light of applicants' disclosure.

Further, the Office has failed to give any probative weight to the advantages and benefits of the present invention as part of the "invention as a whole" and instead has cited references that do **not in any way** disclose or teach such advantages. None of the references recognize that by inactivating the transformation abilities of E6 or E7 proteins there is a reduced likelihood of tumor formation, however, the transformation deficient protein will still trigger an immune response.

In light of the above discussion and the fact that each and every recited limitation of applicants' claimed invention is not disclosed or suggested in the cited references, it is clear that the cited combination fails to establish a *prima facie* case of obviousness of applicants' claims as herein amended.

Claims 16, 18, 20 and 50 stand rejected under 35 U.S.C. §103 (a) as being unpatentable over Donnelly, et al., Johnson, and Whittle, et al. in further view of Gissmann, et al.; and claim 61 over the first three previous references in further view of Stanley, et al. Regardless of the teachings of Gissmann, et al. or Stanley, et al. applicants respectfully submit that the defects in the alleged *prima facie* case over Donnelly, et al., Johnson and Whittle, et al. are not cured by the addition of Gissmann, et al. or Stanley, et al.

In light of the reasons set forth above and the clarifying amendments to the claims, applicants submit that the cited references fail to suggest the subject matter of the currently amended claims. Reconsideration and withdrawal of the rejections under 35 U.S.C. §103 (a) is respectfully requested.

Conclusion

Applicants have satisfied the requirements for patentability. All pending claims are free of the art and fully comply with the requirements of 35 U.S.C. §112. It therefore is requested that Examiner Woitach reconsider the patentability of claims 14-61, 65-66, in light of the distinguishing remarks herein and withdraw all rejections, thereby placing the application in condition for allowance. Notice of the same is earnestly solicited. In the event that any issues remain, Examiner Woitach is requested to contact the undersigned attorney at (919) 419-9350 to resolve same.

Respectfully submitted,

Marianne Fuierer
Registration No. 39,983
Attorney for Applicant

INTELLECTUAL PROPERTY/
TECHNOLOGY LAW
Telephone: (919) 419-9350
Fax: (919) 419-9354
Attorney Ref: 4121-107 RCE 2

APPENDIX A

DAVIES,
RUBERY,
HERALL

THE ENCYCLOPEDIA OF Molecular Biology

EDITOR IN CHIEF
SIR JOHN KENDREW

EXECUTIVE EDITOR
ELEANOR LAWRENCE

b
Blackwell
Science

Blackwell Science Ltd
Editorial Offices:
Osney Mead, Oxford OX2 0EL
25 John Street, London WC1N 2BL
23 Ainslie Place, Edinburgh EH3 6AJ
238 Main Street, Cambridge,
Massachusetts 02142, USA
54 University Street, Carlton,
Victoria 3053, Australia

Other Editorial Offices:
Arnette Blackwell SA
1, rue de Lille, 75007 Paris
France

Blackwell Wissenschafts-Verlag GmbH
Kurfürstendamm 57
10707 Berlin, Germany

Feldgasse 13, A-1238 Wien
Austria

First published 1994
Reprinted as a paperback 1995

Set by Semantic Graphics, Singapore
Printed in Great Britain at the Alden Press Limited,
Oxford and Northampton
and bound by Hartnolls Ltd, Bodmin, Cornwall

DISTRIBUTORS

Marston Book Services Ltd
PO Box 87
Oxford OX2 0DT
(*Orders* Tel: 01865 791155
Fax: 01865 791927
Telex: 837515)

USA
Blackwell Science, Inc.
238 Main Street
Cambridge, MA 02142
(*Orders* Tel: 800 215-1000
617 876-7000
Fax: 617 492-5263)

Canada
Oxford University Press
70 Wynford Drive
Don Mills
Ontario M3C 1J9
(*Orders* Tel: 416 441-2941)

Australia
Blackwell Science Pty Ltd
54 University Street
Carlton, Victoria 3053
(*Orders* Tel: 03 347-0300
Fax: 03 349-3016)

A catalogue record for this title
is available from both the British Library
and the Library of Congress

ISBN 0-632-02182-9
0-86542-621-X (pbk)

full transforming potential, which interacts with the tumour suppressor protein p53 [5].

Polyoma virus LT shares some functions with its SV40 counterpart. It probably has a similar role in replication: DNA binding and ATP-dependent helicase activity have been demonstrated, and it is also active in transcriptional activation/repression assays. It is less effective, however, in transformation assays and is capable only of promoting proliferation, or IMMORTALIZATION of primary tissue culture cells. Interaction with p105Rb has been shown, but no interaction with p53 has yet been demonstrated.

MT. Mouse and hamster polyomaviruses, but not SV40-type viruses, encode a 55K major early function. The hamster species is relatively poorly characterized, but mouse polyoma virus MT has been much studied. It is responsible for cellular alterations which lead to transformation of proliferating cells to a tumorigenic phenotype. MT is located in higher order structures in the cytoplasm, predominantly membrane and cytoskeletal in nature. In particular, a membrane-bound subpopulation associated with plasma membranes, essential to transformation, interacts with and activates the product of the cellular proto-oncogene *c-src* (*see* ONCOGENES). This complex is also directly associated with the 81K component of a phosphatidylinositol-3 kinase enzyme, which may also be found in association with activated platelet-derived growth factor receptor (*see* GROWTH FACTORS; GROWTH FACTOR RECEPTORS). In addition, MT interacts with the 60K, regulatory (A), and the 35K, catalytic (C), subunits of the cellular protein phosphatase 2A complex. The function of this interaction in transformation by MT is unknown.

The relevance of these interactions to the growth of the virus is similarly unclear. MT is necessary for lytic growth: mutations in the MT/ST unique region result in viruses that grow poorly. There is some correlation between this loss of function and correct post-translational modification of the major capsid protein, VP1, which results in a block in virion assembly.

ST. The small T gene product (23K in size) shares common sequence with the other early antigens of the polyomaviruses for most of its length; in mouse polyoma virus only the C-terminal four amino acids are unique to ST. It is dispensable for achieving most viral functions, but it appears to potentiate the actions of the other early antigens and may have a role in tumour formation *in vivo*. STs from both SV40 and polyoma virus interact with the A and C subunits of protein phosphatase 2A, which localizes the site of this interaction to a cysteine-repeat region common to polyoma virus MT and ST and SV40 ST.

The late region

This region encodes the three structural proteins of the virion. The major coat protein, VP1 (45K), is expressed at late times in the viral life cycle and is initially located in the nucleus. Later, it can be found in the cytoplasm of infected cells as well. In mouse polyoma virus it is modified post-translationally by phosphorylation and acetylation and has six isoelectric forms. It can spontaneously organize, *in vitro*, into aggregates of five molecules known as capsomeres, which, under the appropriate buffer conditions, can form capsids. Despite the capsomers being arranged in the capsid with axes of five- and sixfold symmetry (Fig. P3), only pentamers of VP1 are found.

The role of the minor coat proteins in the virion structure is less well defined. X-RAY CRYSTALLOGRAPHIC data have revealed that below the outer layer of VP1 capsomeres lies an electron-dense layer, probably composed of VP2 and 3 (35 and 23K, respectively). VP3 is entirely encoded within VP2 and so its contribution to viral infectivity is difficult to assess, although it is thought to be necessary for efficient viral reproduction. Both proteins accumulate in the nucleus and may be involved in transporting VP1 to this site. VP2 is modified post-translationally with myristic acid and loss of this modification reduces, but does not abolish, infectivity.

Papillomaviruses

It has not been possible to develop an *in vitro* lytic system for the study of papillomaviruses: virus and viral transcripts can only be obtained in very low amounts from infected epithelial tissues or papillomas. However, mRNAs isolated from these sources reveal overlapping and spliced transcripts, and promoters and POLYADENYLATION sites utilized vary, depending on the source and open reading frame (ORF) transcribed. Expression of the viral genes is tightly restricted by the level of differentiation of the cell in the epidermis. With the progression from undifferentiated basal layer cells to differentiated squamous cells, viral gene expression switches from early to late. Viral functions have mostly been analysed by cloning the ORFs into suitable vectors and expressing them in tissue culture. Association of some types of human papillomaviruses (HPVs) with cancerous conditions has stimulated a wealth of data on the functions encoded by these

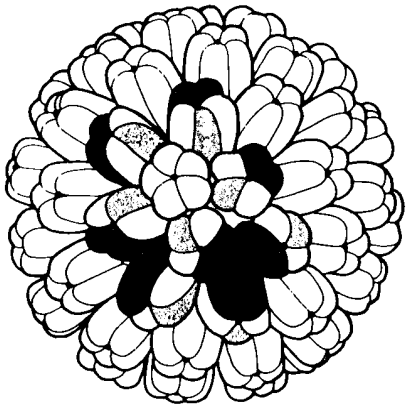
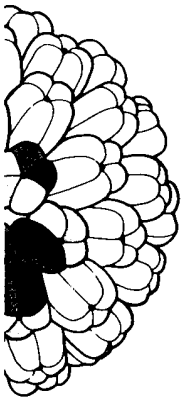


Fig. P3 The packing arrangement of subunits in the papovavirus SV40. The crystal structure of SV40 revealed a new type of packing in which 72 pentamers of virion protein VP1 are arranged in a pattern of interlocking pentagons and hexagons. Twelve 5-coordinated pentamers lie on the 5-fold rotation axes of the icosahedron, each surrounded by five pentamers. Sixty 6-coordinated pentamers do not lie on any symmetry axes and each is surrounded by six pentamers. Based on [7].

ıppropriate buffer conditions, omers being arranged in the old symmetry (Fig. P3), only

s in the virion structure is less PHIC data have revealed that omeres lies an electron-dense and 3 (35 and 23K, respec- m VP2 and so its contribution s, although it is thought to be ction. Both proteins accumu- olved in transporting VP1 to slationally with myristic acid uces, but does not abolish,

an *in vitro* lytic system for the l viral transcripts can only be ı infected epithelial tissues or ted from these sources reveal s, and promoters and POLY- lepending on the source and ibed. Expression of the viral el of differentiation of the cell ession from undifferentiated squamous cells, viral gene o late. Viral functions have he ORFs into suitable vectors ire. Association of some types with cancerous conditions has : functions encoded by these



nits in the papovavirus SV40. The ype of packing in which 72 ged in a pattern of interlocking inated pentamers lie on the 5-fold rounded by five pentamers. Sixty symmetry axes and each is ']

viruses. However, bovine papillomavirus, type 1 (BPV-1) being more readily obtained than other members of the family, is commonly used as a prototype for studying viral functions. As with polyomaviruses, the genome has been divided into early and late regions (Fig. P2), although, unlike polyomaviruses, transcription of both regions occurs from a single strand.

Early region

The region containing early ORFs has been defined as the fragment of the genome (69% of the total) necessary for conferring transformation on cells in an *in vitro* assay. This region contains eight ORFs, five of which (E1, E2, E4, E6, and E7) appear to be found in other papillomavirus genomes, suggesting that these are essential functions.

E1. The E1 ORF gene is involved in episomal (extrachromosomal) replication of the viral genome. It bears some homology with SV40 LT in the regions involved in ATPase activity and nucleotide binding and its product can bind DNA, either alone, or complexed to E2 gene products.

E2. These polypeptides have the properties of transcriptional regulators [6]. They bind as dimers to the palindromic DNA sequence ACCGNNNNCGGT, which is found in several copies in the noncoding region of the virus. Simultaneous binding of E2 dimers to several of these sites results in a high level of transcriptional activation. There is also evidence for cooperation of E2 with cellular transcription factors such as AP1. Active E2 seems to repress promotor/enhancer activity for the ORFs E6 and E7. E2 is often found to be defective in material derived from HPV tumours.

E6, E7. The major proteins encoded by papillomaviruses that are relevant to transformation are transcribed from the E6 and E7 ORFs. These polypeptides are capable of transforming primary cells in a classical focus formation assay. The E6 protein, from HPV 16 and 18, can promote the breakdown of the tumour suppressor protein p53, in an ATP-dependent manner and utilizing the ubiquitin degradation pathway (*see* PROTEIN DEGRADATION). E7, which cooperates with the oncogene *ras* to transform primary cells, has been shown to interact with the underphosphorylated form of the p105Rb tumour suppressor gene. This interaction probably occurs through the N-terminal 37 amino acids, which share some homology with domains 1 and 2 of adenovirus E1A, also known to be involved with p105Rb binding. Therefore, transformation of cells by E6 and E7 may occur by a similar route to that of the DNA tumour viruses — adenovirus and SV40. E6 and E7 may also induce host chromosomal alterations.

E5. A further contribution to transformation, in some papillomaviruses, may come from the product of the E5 ORF. Studies on the 44 amino acid product of deer or bovine papillomavirus E5 ORFs have revealed a potential for the gene in inducing growth, cellular transformation, DNA synthesis, and activation of the platelet-derived growth factor receptor. Various mechanisms for the latter have been proposed, including direct binding to the receptor. Some homology with the β chain of platelet-derived growth factor (*see* GROWTH FACTORS) has been noted. The E5 protein has been isolated in association with a 16K protein thought to be important in cellular compartments in which processing of growth factor receptors occurs.

Late region

The late region has two large ORFs capable of coding for proteins of 55K (L1) and 50K (L2). L1 encodes the major structural capsid protein. The role of the minor capsid protein, L2, is not known.

N.S. KRAUZEWICZ

See also: ANIMAL VIRUSES; ANIMAL VIRUS DISEASE; EUKARYOTIC GENE EXPRESSION.

1 Salzman, N.P. & Howley, P.M. (Eds) (1987) *The Papovaviridae*, Vols 1 and 2 (Plenum, New York).
2 Doerfler, W. & Böhm, P. (Eds) (1992) *Malignant Transformation by DNA Viruses*, 1–85 (VCH, Cambridge).
3 Jones, N.C. et al. (1988) *Trans*-acting protein factors and the regulation of eukaryotic transcription: lessons from studies on DNA tumor viruses. *Genes Devel.* **2**, 267–281.
4 Pipas, J.M. (1992) Common and unique features of T antigens encoded by the polyomavirus group. *J. Virol.* **66**, 3979–3985.
5 Fanning, E. (1992) Simian virus 40 large T antigen: the puzzle, the pieces, and the emerging picture. *J. Virol.* **66**, 1289–1293.
6 McBride, A.A. et al. (1991) The papillomavirus E2 regulating protein. *J. Biol Chem.* **266**, 18411–18414.
7 Liddington, R.C. et al. (1991) Structure of simian virus 40 at 3.8-Å resolution. *Nature* **354**, 278–284.

paracentric inversion *See*: CHROMOSOME ABERRATIONS.

paracrine Term applied to the action of local chemical mediators, which have their effects very close to the cells that secrete them.

paralogous *See*: HOMOLOGY.

paramylon A storage β1,3 glucan of the unicellular alga *Euglena gracilis*, formed by transfer from UDP-Glc, using a paramylon synthetase that is membrane-associated and solubilized by sodium deoxycholate. There is evidence for a GLYCOPROTEIN primer and the synthetase closely resembles CALLOSE synthetase in being stimulated by oligosaccharides of β1,3 and β1,4 glucans.

Paramyxoviridae Family of enveloped RNA ANIMAL VIRUSES comprising three genera — paramyxoviruses, morbiliviruses (measles, canine distemper), and pneumoviruses (respiratory syncytial virus, RSV). Virus particles are pleomorphic and ~150 nm in diameter, with surface spikes. The single-stranded RNA genome (M_r 5×10^6–7×10^6) is usually (−) sense, but some particles contain (+)-sense genomes. *See*: ANIMAL VIRUS DISEASES; ANIMAL VIRUSES.

paranemin An INTERMEDIATE FILAMENT protein.

paraquat One of the low-potential (E_m = −400 mV) viologen dye

General Search Results--Full Record

Article 219 of 340 PREVIOUS NEXT SUMMARY HOLDINGS FIND RELATED RECORDS Explanation
MARK

HUMAN PAPILLOMAVIRUS E6 **AND** E7 - **PROTEINS WHICH DEREGULATE THE CELL-CYCLE**

TOMMASINO M, CRAWFORD L

BIOESSAYS
17 (6): 509-518 JUN 1995

Document type: Review **Language:** English **Cited References:** 62 **Times Cited:** 77

Abstract:
Numerous clinical, epidemiological and molecular findings link some types of Human Papillomaviruses (**HPV**) with cancer of the genital tract, They share a common pathway of **transformation** with a number of DNA tumour viruses, such as Adenovirus and SV40. Although all these viruses are termed 'DNA tumour viruses' and have similar in vitro **transforming** activities, Human **Papillomavirus** is the only one so far clearly involved in human cancer. Extensive studies on **HPV E6** and **E7** proteins have demonstrated their involvement in malignant **transformation**. **E6** and **E7** bind the products of tumour suppressor genes, p53 and Rb1, respectively, modifying or inactivating their normal functions, The Rb1 and p53 genes are deleted or mutated in several cancers and both proteins regulate the transcription of genes involved in cell cycle progression control. The **E6**/p53 and **E7**/Rb1 interactions result in a deregulation of the cell cycle with loss of control of crucial cellular events, such as DNA replication, DNA repair and apoptosis.

KeyWords Plus:
RETINOBLASTOMA GENE-PRODUCT, TYPE-16 **E7**, HUMAN KERATINOCYTES, COMPLEX-FORMATION, ZINC-BINDING, EJ-RAS, P53, **TRANSFORMATION**, ASSOCIATION, ONCOPROTEIN

Addresses:
TOMMASINO M, UNIV CAMBRIDGE,DEPT PATHOL,IMPERIAL CANC RES FUND,TUMOUR VIRUS GRP,TENNIS COURT RD,CAMBRIDGE CB2 1QP,ENGLAND

Publisher:
COMPANY OF BIOLOGISTS LTD, CAMBRIDGE

IDS Number:
RE636

ISSN:
0265-9247

Article 219 of 340 PREVIOUS NEXT SUMMARY

THE EXPRESSION LEVELS OF THE HUMAN PAPILLOMAVIRUS TYPE-16 E7 CORRELATE WITH ITS TRANSFORMING POTENTIAL

LIU ZJ, GHAI J, OSTROW RS, FARAS AJ

VIROLOGY
207 (1): 260-270 FEB 20 1995

Document type: Note **Language:** English **Cited References:** 66 **Times Cited:** 11

Abstract:
The **transforming** potential of the human **papillomavirus** (**HPV**) type 16 has been defined largely in the **E7**, **E6**, and E5 oncoproteins, with the major **transforming** capability residing in the **E7** gene. In this paper, we found that in cooperation with the activated res the HPV16 **E7** gene when expressed in a retroviral vector could fully **transform** baby rat kidney (BRK) cells in transfections, whereas the same construct could only immortalize the BRK cells following retroviral infection. This inability to **transform** correlated with the low levels of **E7** gene RNA expression in the viral infected cells, which harbor a lower number of copies of the **E7** gene constructs. Cotransfection of the expression vector FV2E7, which gives high levels of **E7** gene expression, and activated ras lead to rapid and efficient morphological **transformation** of BRK cells which grew easily in soft agar and induced large tumors in athymic nude mice. In contrast, cotransfections of the expression vector FV1E7, which gives lower levels of 67 gene expression, produced much lower numbers of **transformed** colonies which took longer to form, showed a retarded growth on soft agar, and induced smaller tumors in nude mice. Under these conditions, colonies of immortalized, but morphologically untransformed cells formed in large numbers. These results indicate that the **transforming** potential is directly correlated to the expression levels of the oncoprotein and that a threshold level of the **E7** oncoprotein may be required before the cells can be fully **transformed**. This supports the hypothesis that the **transformation** processes include at least two separate and continuous steps which first lead to immortalization and then to metastasis, in agreement with the clinical progression of genital tumors from benign to malignancy. Such a progression may involve enhanced expression of the oncoproteins. (C) 1995 Academic Press, Inc.

KeyWords Plus:
CARCINOMA CELL-LINES, OPEN READING FRAME, CERVICAL-CARCINOMA, ONCOGENIC **TRANSFORMATION**, HUMAN KERATINOCYTES, ACTIVATED RAS, GENE-PRODUCTS, **E6** PROTEIN, DNA, IMMORTALIZATION

Addresses:
UNIV MINNESOTA,SCH MED,INST HUMAN GENET,MINNEAPOLIS,MN 55455
UNIV MINNESOTA,SCH MED,DEPT MICROBIOL,MINNEAPOLIS,MN 55455

Publisher:
ACADEMIC PRESS INC JNL-COMP SUBSCRIPTIONS, SAN DIEGO

IDS Number:
QJ657